

ABSTRACT

The present invention is directed to an improved multiplex PCR method for obtaining at least two PCR products from one PCR solution. In the multiplex PCR method for having at least two DNA amplified products from a 5 sample positioned in a PCR equipment, the object of the present invention is to provide a novel multiplex PCR method characterized in that a primer annealing temperature and an extension time be changed per cycle with constant periods. When a multiplex PCR is performed in accordance with the present invention, limitations in determining PCR conditions due to a 10 various sizes of PCR products or dimers caused by primers can be eliminated, and time and efforts required for determining the PCR conditions to various samples can also be reduced. Further, not only refined DNAs like cDNA, genomic DNA, vector, etc, but blood can be directly used as a multiplex PCR sample, and PCR amplification reaction can be performed even with a sample 15 having the smallest amounts.